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Epidemiological consequences of a pathogen having both virulent and avirulent modes of transmission: the case of rabbit haemorrhagic disease virus

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SUMMARY

A number of pathogens cause chronic infection in survivors of acute disease and this is believed to be a common means of persistence, including for highly virulent agents. We present a model in which transmission from chronically infected hosts causes *chronic* infection in naive individuals, without causing acute disease – indeed 'protecting' against it. Thus the pathogen obtains the benefit of virulence (high transmission rate), but mitigates against the cost (high host mortality). Recent findings suggest that rabbit haemorrhagic disease virus (RHDV), a highly contagious and virulent pathogen, may also utilize this alternative, 'avirulent', mode of transmission. The model may resolve the paradox of how RHDV can be highly prevalent in some populations, in the absence of mortality. Differences in host demography determine whether avirulent transmission prevents large-scale mortality (as in most UK populations) or not. Other pathogens may exhibit similar behaviour and the implications for emerging diseases in general are discussed.

INTRODUCTION

In the typical course of infection of a competent host, the infecting pathogen multiplies and causes the host to become infectious. The level of infection increases until a point of crisis is reached, when either the host dies, or develops immunity and infectiousness ceases. However a number of pathogens are able to remain active in the host even after immunity has developed, causing a long-term chronic infection with the pathogen shed at reduced rates (e.g. *Salmonella typhi*, *Mycobacterium tuberculosis* and *M. bovis*, hepatitis B virus, herpes simplex virus). Such an infectious chronic state can be lifelong. A number of models featuring infectious chronic infection have been developed, in which the outcome of a transmission event is determined by the status of the 'recipient' of infection, with all infections of naive hosts being acute (e.g. [1, 2]). However, the status of the *source* of infection may also be important, with the quantity of pathogen shed determining the type of infection (acute or chronic) developed by the recipient. In the model presented here, the nature of new infections depends upon the status of the source of infection, not the recipient. Thus the model has the novel feature that the infectious agent has two modes of transmission that are simultaneously in 'competition' for susceptibles. In light of new evidence for an avirulent mode of transmission of the highly virulent rabbit haemorrhagic disease virus, we apply the model to examine the paradox of how a pathogen that causes huge mortality in some populations apparently persists in others at high prevalence in the absence of disease. Additionally we examine the advantage to the pathogen of having two phases of infection.

Rabbit haemorrhagic disease virus (RHDV) is a highly virulent pathogen that kills up to 95% of infected rabbits (*Oryctolagus cuniculus*) 48 h post-infection [3–6]. Outbreaks killed 140 million farmed

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rabbits in China in 1984 [7], 64 million farmed rabbits in Italy in 1986 [8] and 30 million wild rabbits in Australia in just a few weeks following its release in 1995 [6]. However, immunity to RHDV has been found in the absence of signs of disease in both wild and captive populations from a number of locations, including the former Czechoslovakia, Switzerland, Austria, Germany and Sweden [9–13]. Seroprevalence was particularly high in the United Kingdom, with a mean of 64% and a maximum of 100% [14, 15]. It has been proposed that this immunity may be due to a non-pathogenic strain of RHDV [10–14], such as that isolated from a rabbitry in Italy in 1996 [9, 16].

However, despite extensive and intensive sampling this non-pathogenic strain has not been found elsewhere, and recent research by Moss et al. [17] has suggested that the same virus can be responsible for both large-scale mortality and also highly-prevalent avirulent infection, found in the absence of mortality. Moss et al. [17] used RT–PCR to test >40 serum samples from healthy UK rabbits, both captive and wild, from five locations, that showed no signs of disease, either at the time of sampling or in subsequent weeks and months. Half of the samples contained detectable levels of RHDV, but in no case was the 'Italian' non-pathogenic strain detected. Nucleotide sequences were indistinguishable from those of pathogenic RHDV by phylogenetic analysis of 527 nucleotides that encode part of the outer region of the VP60 capsid protein. This region of the genome has been used in most phylogenetic studies of RHDV (e.g. [11]), and contains significant differences between pathogenic RHDV and the 'Italian' non-pathogenic strain [9, 16]. In all cases, sera that contained RHDV also contained antibodies against the virus, suggesting that long-term infection had occurred, despite a host immune response. Consistent with this, Shien et al. [18] reported that virus was detectable for at least 47 days (the end of the experiment) in survivors of acute (experimental) infection. The aforementioned finding of very high seroprevalence (up to 100%) in the absence of mortality strongly suggests that both infection and subsequent transmission can occur in the absence of acute disease. Furthermore, RHDV was detected in rabbit serum samples taken in the 1950s, which implies that the virus has been circulating in the United Kingdom for at least 40 years before it was detected. This complements and extends the findings of Rodak et al. [13] who reported the presence of anti-RHDV antibodies in Czech rabbit sera collected in 1975, a decade before the discovery of RHDV.

There is a marked difference in the levels of virus in acute and chronically infected rabbits, which may explain the different modes of transmission. In acute-infected rabbits, RHDV is found at very high titres, being detectable in liver homogenate even after 10^9-fold dilution [5], and flyspots can contain enough RHDV to cause acute infection, indicating that it is shed at high concentration [19]. In contrast, detection of the virus in the sera of healthy rabbits required the use of nested RT–PCR to enhance sensitivity [17], and virus was found at low levels in survivors of experimental infection [18]. Note that such low viral titres are not incompatible with successful transmission of RHDV: detection of the 'Italian' non-pathogenic strain also requires enhanced-sensitivity techniques, yet it was shown to be transmissible, and indeed it persisted in a rabbitry for at least 2 years [9, 16].

The determinant of whether the initial infection with RHDV results in acute disease or chronic infection may be the viral 'dosage' received – which may vary by several orders of magnitude, considering the evidence above. Following infection, virus replicates at a rapidly accelerating rate. The antibody-mediated immune response occurs 2–3 days post-infection [3] and if this immune response succeeds in arresting viral amplification before a fatal amount of liver damage occurs then the rabbit survives and develops chronic infection, otherwise death results. We expect that the time taken for fatal damage to occur will depend upon the size of the initial viral inoculum, with acute infection resulting from a 'large' inoculum (received from an acute-infected rabbit) enabling infection to progress rapidly, usually overwhelming the host before antibodies have been produced in sufficient quantity to control the infection. A smaller inoculum (from a chronically infected rabbit) would allow more time for the host immune system to respond before fatal liver damage occurs. Thus the two phases of infection may be simply a consequence of the kinetics of infection and the host's immune response, without requiring any specialized 'molecular machinery' on the part of the virus to effect a 'switch' in behaviour. In general support of this conjecture regarding inoculum-dosage-dependent effects, Timms et al. [20] showed experimentally with malaria in mice that both disease severity and rate of progression increased with the size of the inoculum.

In this paper we use a modelling approach to examine the epidemiological consequences of the following hypothetical scenario, which is illustrated in Figure 1. Acute RHDV infection is short-term, with

high disease-associated mortality, is highly infectious and causes acute infection upon transmission, due to a high rate of viral shedding. Survivors of acute infection develop chronic infection, which is longer-term, much less infectious and causes chronic infection upon transmission, due to a low rate of viral shedding. Susceptible rabbits that acquire RHDV from a chronic-infected rabbit develop chronic infection themselves, without experiencing the acute phase. Chronic infection does not cause any disease-associated mortality.

We examine whether this model can explain the intriguing situation in the United Kingdom, where immunity to RHDV, in the absence of observable disease, was found in all 68 of the wild rabbit populations sampled, usually at very high prevalence (mean 64%, range 10–100%) [14, 15]. Furthermore, in the United Kingdom, virulent RHDV has had little impact, nationally, in terms of the number of populations affected and the total number of rabbits that died, despite scattered lethal outbreaks having occurred in wild populations country-wide since 1994 [21; P. J. White unpublished observations]. It appears that there may be herd immunity, due to chronic infection, protecting most UK wild rabbit populations. Consistent with this, there is a geographical trend of mean seroprevalence decreasing from north to south, and it is in the south where the majority of lethal outbreaks have occurred. Furthermore, most UK populations had much higher seroprevalence than mainland European populations (where 12, 19 and 46% have been reported [12, 22]), which may explain the much lesser impact of RHDV in the United Kingdom compared to other infected countries.

Can the model explain how RHDV may have persisted in the United Kingdom, unnoticed, at high prevalence, for decades before lethal outbreaks were recorded from the 1990s? Can it explain why seroprevalence is so variable and has a trend of increasing from south to north? Furthermore, can it explain why the United Kingdom has had only a few lethal outbreaks, whereas other countries have been affected much more severely? Finally, we examine how the virus may benefit from having two phases of infection.

MODEL ANALYSIS

Description

A flow diagram representing the model is shown in Figure 1, with parameters summarized in Table 1.

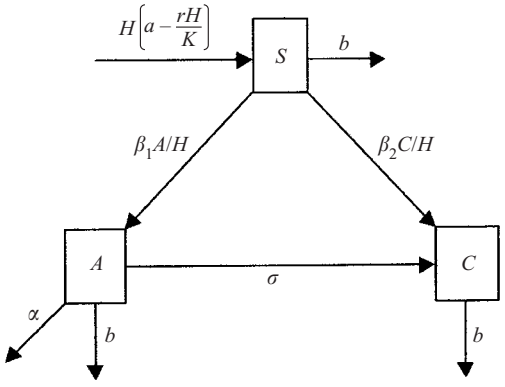

Fig. 1. Flow diagram of the model. Susceptible rabbits (S) may acquire acute infection from an acute-infected individual (A) at rate β_1, or chronic infection from a chronic-infected individual (C) at rate β_2. Rabbits with acute infection are subject to the disease-induced death rate, α, and those that recover, at rate σ, develop chronic infection. All individuals are subject to the natural death rate, b. There is density dependence in the productivity of the population. Parameter estimates are in Table 1.

The equations are as follows,

$$H = S + A + C,$$

$$d = \alpha + \sigma + b,$$

$$r = a - b,$$

$$\frac{\mathrm{d}H}{\mathrm{d}t} = rH\left(1 - \frac{H}{K}\right) - \alpha A,$$

$$\frac{\mathrm{d}S}{\mathrm{d}t} = H\left(a - \frac{rH}{K}\right) - \frac{S}{H}(\beta_1 A + \beta_2 C) - bS,$$

$$\frac{\mathrm{d}A}{\mathrm{d}t} = \beta_1 S\frac{A}{H} - \mathrm{d}A,$$

$$\frac{\mathrm{d}C}{\mathrm{d}t} = \beta_2 S\frac{C}{H} + \sigma A - bC.$$

Population dynamics parameters were estimated from published data [23–27], as described in Appendix (a). The productivity rate (the product of the birth rate and nestling survival rate) is density-dependent, reflecting the findings of Myers et al. [25] and Thompson [26]. They reported that the maximum productivity rate was double the rate at equilibrium, so we take $a = 2b$, for all values of b used in this paper.

Within rabbit populations, RHDV is probably transmitted by direct contact [28], through the respiratory route, since the virus can be detected in the airways of infected rabbits, which rapidly infect cage-mates and experimental infection can be caused by intranasal inoculation [16, 28, 29]. Rabbits are a social

Table 1. *Parameter definitions and estimates. Rates are* per capita *per day*

a	Maximum productivity rate (in this paper, $a=2b$ for all values of b)	0·00578–0·0342
b	Natural death rate	0·00289–0·0171
K	Carrying capacity	100
α	Disease-induced death rate (due to acute infection)	0·475
σ	Rate of recovery from acute infection	0·025
d	Rate of loss of acute-infected individuals	0·50289–0·5171
β_1	Transmission parameter (β_1: acute, β_2: chronic)	$\beta_1=0{\cdot}936$; $\beta_2=0{\cdot}0239$

species, making the rate at which each individual contacts others insensitive to changes in total population size. Therefore we assume that the rate of disease transmission will be frequency dependent, which is supported by empirical studies [30, 31].

Acute infection typically lasts for 2 days, with 95% mortality amongst infected rabbits [3–6]. Thus $(\alpha+\sigma)=0{\cdot}5$ and $\alpha/(\alpha+\sigma)=0{\cdot}95$; so $\alpha=0{\cdot}475$; $\sigma=0{\cdot}025$. In the model, chronic infection does not induce mortality and since virus has always been found circulating in the presence of antibodies [17] we postulate that infectiousness is lifelong. Each phase has its own basic reproductive ratio, R_0, for the acute phase, $R_{0,A}=\beta_1/d$ and for the chronic, $R_{0,C}=\beta_2/b$ [see Appendix (*b*)].

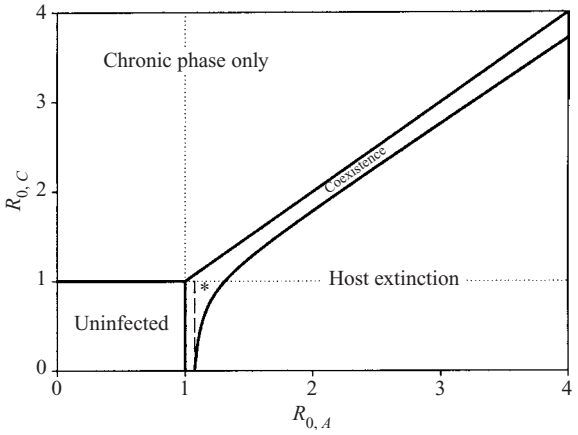

Fig. 2. R_0 map, showing the effects on equilibrium behaviour of varying $R_{0,A}$ and $R_{0,C}$, by changing β_1 and β_2, respectively. There are four modes: uninfected ($R_{0,A}$, $R_{0,C}<1$); chronic phase infection only ($R_{0,C}>1$, $R_{0,A}<R_{0,C}$); coexistence:

$$R_{0,A}>1,\ R_{0,A}>R_{0,C}>\frac{R_{0,A}[rR_{0,A}(b+\sigma)-\alpha b(R_{0,A}-1)]}{d[rR_{0,A}-\alpha(R_{0,A}-1)]}$$

and host extinction:

$$R_{0,A}>1,\ R_{0,A}>\frac{R_{0,A}[rR_{0,A}(b+\sigma)-\alpha b(R_{0,A}-1)]}{d[rR_{0,A}-\alpha(R_{0,A}-1)]}>R_{0,C}.$$

The uninfected equilibrium is relevant for all values of $R_{0,A}$ and $R_{0,C}$. The chronic-phase-only infected equilibrium (K, 0, C^*), is relevant where $R_{0,C}>1$ but between the horizontal dashed line ($R_{0,C}=1$) and the diagonal line ($R_{0,A}=R_{0,C}$) it can be invaded by the acute phase, moving the system to coexistence (H^*, A^*, C^*) or host extinction. The coexistence and host extinction equilibria can only occur following introduction of acute infection. Notice that in the region (labelled *) bounded by the three lines, $R_{0,C}=1$, $H^*=0$ and the long-dashed line, the virus is able to persist *only* because both phases are present: without the chronic phase, the acute would cause host extinction, whilst the chronic phase alone could not invade the population. Apart from β_1 and β_2, which are varied, other parameter values are as in Table 1, with population demographic parameters corresponding to the UK mean, i.e. $b=0{\cdot}00862$; $a=0{\cdot}01724$; $d=0{\cdot}50862$.

Equilibrium analysis

The model has three infected equilibria (Fig. 2), of the form (H, A, C): (i) with chronic-phase-infected individuals only (K, 0, C^*), (ii) 'coexistence' with both acute and chronic individuals (H^*, A^*, C^*) and (iii) host extinction due to RHDV. The chronic-phase-only equilibrium (K, 0, C^*), is relevant where $R_{0,C}>1$ (i.e. $\beta_2>b$), and stable where $R_{0,A}<R_{0,C}$. Due to competition for susceptibles, where the chronic phase has the higher R_0 it excludes the acute (see [32]). The converse does not apply because chronic phase individuals result from the recovery of acutes. Thus there is no acute-phase-only equilibrium, but rather coexistence, with both phases present, which is relevant where $R_{0,A}>1$ and $R_{0,A}>R_{0,C}$. At the threshold where coexistence (H^*, A^*, C^*) becomes relevant ($R_{0,A}=R_{0,C}$), the chronic-phase-only equilibrium (K, 0, C^*) becomes unstable [Appendix (*c*)], but remains relevant. This means that, where $R_{0,A}>R_{0,C}$, the system may be at (K, 0, C^*) but this can be invaded by acute

infection, moving the system to (H^*, A^*, C^*) or host extinction (see below).

Chronic infection does not cause disease-induced mortality, so in the chronic-phase-only equilibrium, the population is at carrying capacity. Where there is coexistence, disease-induced mortality due to acute infection depresses the population below carrying capacity. Indeed, host extinction can occur if acute infection is sufficiently prevalent. The boundary for host extinction occurs where $H^*=0$, and in $R_{0,A}-R_{0,C}$ space it is relevant where $R_{0,A}>1$ and $R_{0,C}>0$. Expressions for H^* and the boundary $H^*=0$ in $R_{0,A}-R_{0,C}$ space, respectively, are:

$$H^* = K\left[1 - \frac{\alpha(b\beta_1 - d\beta_2)(\beta_1 - d)}{r\beta_1[\beta_1(b+\sigma) - d\beta_2]}\right];$$

$$R_{0,C} = \frac{R_{0,A}[rR_{0,A}(b+\sigma) - \alpha b(R_{0,A}-1)]}{d[rR_{0,A} - \alpha(R_{0,A}-1)]}.$$

The likelihood of host extinction is greatly reduced by the infectious chronic phase (Fig. 2). If $R_{0,C}=0$ then the model becomes SIR-type, and examination of the horizontal axis of Figure 2 shows that infected equilibrium is possible only for a small range of $R_{0,A}$ values. Thus the presence of avirulent transmission promotes the persistence of the virus, and indeed the acute phase, by reducing the likelihood of host extinction. Also, it is possible for the chronic phase to persist due to the presence of the acute, when it alone could not invade a naive population. Thus there is a region of parameter space where neither phase alone could persist in a population, but together they can coexist (Fig. 2). Note that the size of the region of coexistence is sensitive to the disease-induced mortality rate, α, increasing as it declines. A pathogen less virulent than RHDV – with a lower case-fatality rate (the proportion of those with acute infection that die of disease), and/or a longer time to death – can have a much larger region of coexistence.

Since the R_0s of the two phases are affected differentially by the natural death rate, b, host population dynamics can affect the behaviour of the system, determining which of the three infected equilibria are stable. The death rate of acute-infected rabbits, d, is insensitive to b (because $\alpha \gg b$); whereas chronic-phase individuals are lost only through natural death. So $R_{0,A}$ is almost invariant, whereas $R_{0,C}$ is very variable among populations with different dynamics. Thus each phase may dominate in some populations and not others, leading us to ask the questions, 'Can this variation in $R_{0,C}$ explain the range of seroprevalence recorded in the United Kingdom in the absence of disease?', and, 'Have most of the UK rabbit populations, particularly those in the north, been protected by the chronic phase infection, in contrast to European populations?'

UK seroprevalence and estimation of transmission parameters

The range of seroprevalence recorded in the absence of disease in the United Kingdom (10–100%) [14, 15] indicates that $R_{0,C}$ has markedly different values in different populations. This large variability in seroprevalence between populations then leads us to ask, 'Can differences in population dynamics explain this range of seroprevalence, and if so then what proportion of UK populations are likely to be protected by endemic chronic-phase infection?' We estimate β_2 (the chronic phase transmission parameter) by assuming that a population with typical UK population demography will have seroprevalence at the mean UK level (64%) due to chronic-phase infection only. Then we examine if the range of seroprevalence recorded in the United Kingdom can be explained by differences in the natural death rate. A typical UK population has natural death rate, $b=0{\cdot}00862$ [see Appendix (a)], and using the relationship $\beta_2 = b/(1-\text{seroprevalence})$ [see Appendix (d)], we estimate $\beta_2 = 0{\cdot}0239$. What range of seroprevalence can be explained by differences in the natural death rate, b, due to population demography? The range of b values estimated for UK rabbit populations is 0·00289–0·0171, predicting a range of seroprevalence of 28–88%, which encompasses the seroprevalence figures recorded for most of the UK populations sampled (52 out of 68), and corresponds to $R_{0,C}=1{\cdot}40$–$8{\cdot}26$.

As usual in studies of emerging diseases field data on the course of an epidemic are limited. We estimate the acute phase transmission parameter, β_1, using the following information. An outbreak occurred at Dawlish Warren in Devon [21], shortly after the seroprevalence survey was performed [14, 15]. Although this site was not sampled, six others in Devon were. Their mean seroprevalence was 45%, with the value for the site nearest to Dawlish Warren being 43%. Since these estimates were similar, we assume they were representative of Dawlish Warren, with the chronic phase being endemic at 45% prevalence. Hence we estimate b for Dawlish Warren to be 0·0131, using the estimate of β_2 (see above) and the relationship, $\beta_2 = b/(1-\text{seroprevalence})$ [Appendix (d)]. The

condition for invasion of the chronic-infected equilibrium by the acute phase is $\beta_1 > d\beta_2/b = 0{\cdot}936$. Although this is formally a minimum estimate of β_1 we expect that it is close to the true value, given that the majority of outbreaks have occurred in regions with relatively low seroprevalence [33]. If β_1 were substantially greater than this estimate then we would expect there to have been more fatal outbreaks elsewhere in the United Kingdom, where mean seroprevalence is higher.

Using the estimates for β_1 and β_2, we examine the equilibrium behaviour of the model over the UK range of natural death rate, b (Fig. 3). The chronic-phase-only equilibrium (K, 0, C^*) is relevant over the entire range of b values, and where $b < d\beta_2/\beta_1$ (i.e. $R_{0,A} < R_{0,C}$) it is the only relevant infected equilibrium, because the chronic phase excludes the acute. Where $b > d\beta_2/\beta_1$ (i.e. $R_{0,A} > R_{0,C}$), both chronic-phase-only (K, 0, C^*) and coexistence (H^*, A^*, C^*) equilibria are relevant. In the chronic-phase-only equilibrium (K, 0, C^*) the population size is at carrying capacity and does not vary with the natural death rate, b, whereas seroprevalence is sensitive to b, declining as it increases. By contrast, in the coexistence equilibrium (H^*, A^*, C^*), the acute phase depresses the population size below carrying capacity and H^* is highly sensitive to b; with host extinction occurring where

$$b > \left[\frac{d\beta_2}{\beta_1} - \frac{r\beta_1\sigma}{r\beta_1 - \alpha(\beta_1 - d)}\right],$$

whereas seroprevalence is insensitive to b. If the chronic phase is endemic in a population where $b > d\beta_2/\beta_1$ (i.e. $R_{0,A} > R_{0,C}$) then acute infection is able to invade, with its introduction moving the system from the chronic-phase-only equilibrium (K, 0, C^*) to the coexistence equilibrium (H^*, A^*, C^*), or causing host extinction. The behaviour of the model reflects the sensitivity of the chronic phase – and insensitivity of the acute phase – to b: as b varies across the range 0·00289–0·0171, the corresponding R_0 ranges are: $R_{0,C}$: 1·40–8·26; $R_{0,A}$: 1·80–1·86. The model predicts that where a population is at the chronic-phase-only (K, 0, C^*) equilibrium, its seroprevalence indicates whether acute infection is able to invade and whether an invasion would lead to depression of the host population, or its extinction (Fig. 3). The lethal outbreaks of RHDV in the United Kingdom occurred where seroprevalence was lowest [33], which is where we suggest natural death rates are highest and thus the chronic-phase-only equilibrium (K, 0, C^*) is able to be invaded by acute infection, because $b > d\beta_2/\beta_1$.

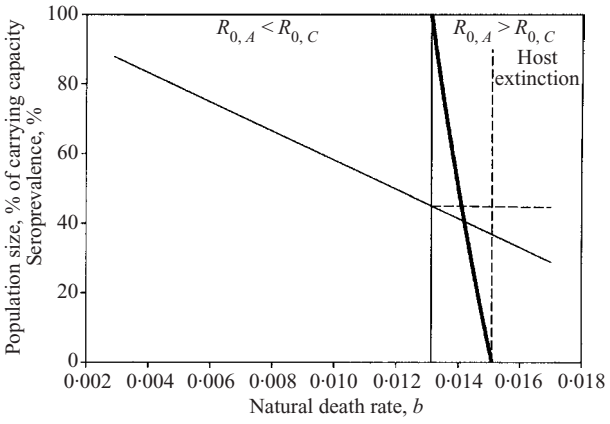

Fig. 3. Effect on equilibrium host population size and seroprevalence of varying the host natural death rate, b, across the UK range. The vertical solid line indicates the threshold $b = d\beta_2/\beta_1$ (i.e. $R_{0,A} = R_{0,C}$). Where $b < d\beta_2/\beta_1$ the only relevant infected equilibrium is chronic-phase-only (K, 0, C^*) (which is relevant over the whole range of b values). Where $b > d\beta_2/\beta_1$ the coexistence equilibrium (H^*, A^*, C^*) is also relevant, and when the acute phase is present, host extinction occurs where

$$b > \left[\frac{d\beta_2}{\beta_1} - \frac{r\beta_1\sigma}{r\beta_1 - \alpha(\beta_1 - d)}\right],$$

to the right of the vertical dashed line. The heavy solid line represents equilibrium population size (expressed as % of carry capacity) when the acute phase is present. (Equilibrium population size when only the chronic phase is present is always 100 % and so is not shown.) The other lines represent seroprevalence, with the solid line indicating seroprevalence in the chronic-phase-only (K, 0, C^*) equilibrium and the dashed line, seroprevalence in the coexistence equilibrium, (H^*, A^*, C^*). Since both the equilibrium population size and seroprevalence are expressed as percentages, they use the same vertical axis.

Dynamics of acute- and chronic-phase infections

The foregoing equilibrium analysis highlights the cost of acute phase infection, with its high disease-induced death rate (α), which means that chronic infection is required to prevent host extinction (except where $R_{0,A}$ is very low) (Fig. 2). Now we consider the benefit of acute infection, which is its high transmission parameter (β_1), leading to its rapid spread through a naive population. Lipsitch and Nowak [34] showed that, if a more virulent strain of virus has a higher transmission parameter than a less virulent one, then this may confer a transient advantage in the invasion of a naive

(or growing) host population, even if the more virulent strain has the lower R_0 and so will be excluded at equilibrium by the less virulent one. We examine the dynamical behaviour of RHDV's invasion of a naive population and then go on to consider how a wild-type strain may be favoured over a hypothetical mutant strain that causes only chronic infection.

Consider the introduction of a rabbit with acute infection with wild-type RHDV (which causes both phases of infection) into a naive population, at $(K, 0, 0)$. Provided $R_{0,A} > 1$ there will follow a rapid epidemic that kills most rabbits, followed by a decline in the prevalence of acute infection. Survivors, in which the infection has entered the chronic phase, reproduce, and the population begins to recover. Transmission from these chronic phase rabbits recruits susceptibles into the chronic phase, protecting them from acute infection. The system tends towards one of the infected equilibria: $(K, 0, C^*)$, (H^*, A^*, C^*) or host extinction, depending upon the relative values of $R_{0,A}$ and $R_{0,C}$ as discussed above. An example of each outcome is shown in Figure 4. Note that in the case of Figure 4(*a*) the chronic ('endemic') mode of transmission 'takes over' from the initial acute ('epidemic') transmission without any attenuation of the virus or evolution of genetic resistance in the host. Despite its R_0 being lower than that of the chronic phase, resulting in its ultimate exclusion, the acute phase's higher transmission parameter means that it spreads more rapidly in a naive population. Taking the approach of Lipsitch and Nowak [34], we define the variable ρ, the ratio of the prevalences of the acute and chronic phases of infection: $\rho = A/C$. The acute phase is favoured when $\mathrm{d}\rho/\mathrm{d}t$ is positive and the chronic phase is favoured when it is negative. The expression for $\mathrm{d}\rho/\mathrm{d}t$ is,

$$\frac{\mathrm{d}\rho}{\mathrm{d}t} = \rho\left[\frac{S}{H}(\beta_1 - \beta_2) - \alpha - \sigma(1+\rho)\right].$$

The acute phase is favoured when $\beta_1 \gg \beta_2$ and the susceptible proportion (S/H) is high, which occurs in a naive population or one that is growing rapidly through the birth or immigration of new susceptible individuals. As the virus spreads through the population, S/H declines, the acute phase loses the advantage and declines in prevalence with respect to the chronic.

Now we compare the transmission success in a naive population of a wild-type virus ('Strain 1'), which causes both acute and chronic phases of infection, with that of an attenuated mutant ('Strain 2') which causes only chronic infection. The chronic phases of Strain 1 and Strain 2 have identical properties. The model is modified as follows: A is replaced by

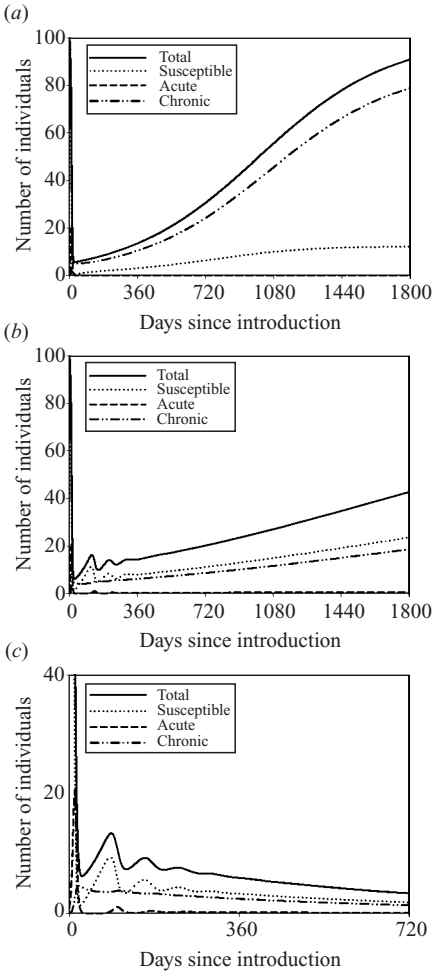

Fig. 4. Effect of introducing a single acute-phase-infected individual into naive populations with different natural death rates. Other parameter values are as in Table 1. Initially the population is in equilibrium $(K, 0, 0)$, at carrying capacity. (*a*) $b < \mathrm{d}\beta_2/\beta_1$ ($b = 0{\cdot}00289$; $a = 0{\cdot}00578$; $d = 0{\cdot}50289$): the acute phase is excluded by the chronic phase, and the population returns to carrying capacity. (*b*) $b > \mathrm{d}\beta_2/\beta_1$ ($b = 0{\cdot}0135$; $a = 0{\cdot}0270$; $d = 0{\cdot}5135$): coexistence is established between the acute and chronic phases and the population is depressed below carrying capacity. (*c*):

$$b > \left[\frac{\mathrm{d}\beta_2}{\beta_1} - \frac{r\beta_1\sigma}{r\beta_1 - \alpha(\beta_1 - d)}\right] (b = 0{\cdot}0171; a = 0{\cdot}0342; d = 0{\cdot}5171),$$

the host population goes extinct ultimately, with both phases coexisting until this moment. Note that the simulation outputs (*a*) and (*b*) are for 5 years (model years are 360 days), whilst (*c*) is for 2 years. Also the vertical axis of (*c*) has a different scale.

A_1; C is replaced by C_1; the expressions for $\mathrm{d}S/\mathrm{d}t$ and H are modified with the inclusion of C_2; and the equation for $\mathrm{d}C_2/\mathrm{d}t$ is added:

$$H = S + A_1 + C_1 + C_2,$$

$$\frac{\mathrm{d}S}{\mathrm{d}t} = H\left(a - \frac{rH}{K}\right) - \frac{S}{H}[\beta_1 A_1 + \beta_2(C_1 + C_2)] - bS,$$

$$\frac{\mathrm{d}C_2}{\mathrm{d}t} = \beta_2 S\frac{C_2}{H} - bC_2.$$

To compare the transmission success of the two strains, we define the variable ρ', the ratio of the prevalences of Strain 1 and Strain 2: $\rho' = (A_1 + C_1)/C_2$. Strain 1 is favoured when $\mathrm{d}\rho'/\mathrm{d}t$ is positive and Strain 2 is favoured when it is negative. The expression for $\mathrm{d}\rho'/\mathrm{d}t$ is,

$$\frac{\mathrm{d}\rho'}{\mathrm{d}t} = x\rho'\left[\frac{S}{H}(\beta_1 - \beta_2) - \alpha\right],$$

where $x = A_1/(A_1 + C_1)$, the proportion of Strain 1 prevalence represented by the acute phase.

Strain 1 is favoured by large x, and a large susceptible proportion (S/H) when $\beta_1 \gg \beta_2$. When invading a naive population (i.e. initial conditions $A_1 = C_2 = 1$, $C_1 = 0$, $S/H \sim 1$), Strain 1 has the advantage when $(\beta_1 - \beta_2) > \alpha$. As invasion proceeds, Strain 1's advantage declines, as both S/H and x decline. However, even if the acute phase (A_1) is excluded ultimately ($\beta_1/d < \beta_2/b$), the prevalence of Strain 1 at equilibrium is greater than Strain 2, even though only their chronic phases (which have identical properties) are present, because of the initial rapid spread of Strain 1, via its acute phase (Fig. 5). Furthermore, where $b > \mathrm{d}\beta_2/\beta_1$, Strain 2 is excluded by the acute phase of Strain 1 (A_1), but the chronic phase of Strain 1 (C_1) coexists (although at high natural death rates, host extinction can occur).

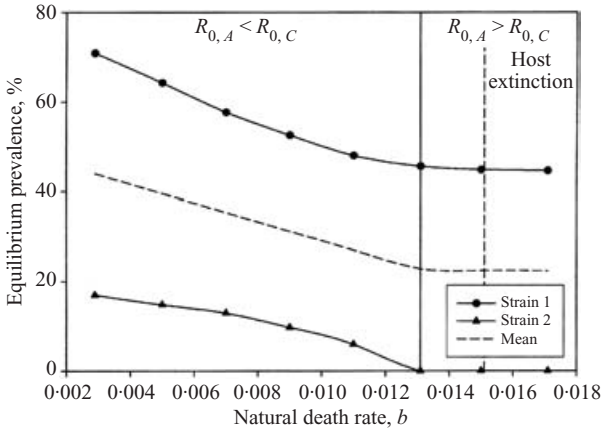

Fig. 5. Equilibrium prevalences of Strain 1 (which has acute and chronic phases) and Strain 2 (chronic phase only) following introduction of one rabbit infected with Strain 1 (in the acute phase) and one rabbit infected with Strain 2 (chronic phase) into a naive population, plotted against natural death rate, b. The vertical solid line indicates the threshold $b = \mathrm{d}\beta_2/\beta_1$ (i.e. $R_{0,A} = R_{0,C}$). Where $b < \mathrm{d}\beta_2/\beta_1$ (i.e. $R_{0,A} < R_{0,C}$), the acute phase of Strain 1 is excluded at equilibrium, leaving only the chronic phases of the two strains. The chronic phases of both strains have identical parameter values, but Strain 1 has higher prevalence due to its more rapid initial spread via its acute phase. (If Strain 1 is introduced in its chronic phase then both strains reach the same, mean, prevalence.) Where $b > \mathrm{d}\beta_2/\beta_1$ (i.e. $R_{0,A} > R_{0,C}$), Strain 2 is excluded by the acute phase of Strain 1, but the chronic phase of Strain 1 is still present due to recovery from acute infection. To the right of the vertical dashed line, which indicates where

$$b = \left[\frac{\mathrm{d}\beta_2}{\beta_1} - \frac{r\beta_1\sigma}{r\beta_1 - \alpha(\beta_1 - d)}\right],$$

acute infection causes host extinction (although, mathematically, Strain 1 still has a prevalence).

DISCUSSION

Theoretical work has shown how pathogen virulence can be adaptive, when it occurs as the result of a trade-off between the transmissibility of the pathogen and the lifetime of the infected host [35]. However, highly virulent pathogens are prone to local extinction, and there are a number of strategies for persistence. Some pathogens utilize alternative 'reservoir' hosts, although none has been found for RHDV, despite extensive testing [5, 36, 37]. Another strategy is to induce a carrier state, in which survivors of acute infection remain infected, shedding the pathogen at reduced rates. In other models featuring infectious chronic infection the outcome of a transmission event is determined by the status of the 'recipient' of infection, with all infections of naive hosts being acute (e.g. [1, 2]). In contrast, in the model presented in this paper, the nature of new infections depends upon the status of the *source* of infection, with its rate of pathogen shedding, and hence the quantity of pathogen transmitted, determining the type of infection (acute or chronic) developed by the recipient. The model has the novel feature that the pathogen has two modes of transmission that are simultaneously in 'competition' for susceptibles.

The model has the important asymmetry that chronic infection can exclude acute, but not vice versa (because survivors of acute infection develop chronic infection). This facilitates persistence of the pathogen at equilibrium, by reducing the likelihood of host extinction. Thus the pathogen is able to enjoy the benefits of virulence, in terms of a high transmission rate, but mitigate against the cost, in terms of high host mortality. It is possible for the same pathogen to cause outbreaks of great mortality and also to persist at high prevalence in the absence of mortality, through transmission of acute and chronic infections, respectively.

To investigate the dynamics of infection with such a pathogen, we applied the model to RHDV, following the important new findings of Moss et al. [17], that the same virus may cause both large-scale mortality, and also avirulent infection at high prevalence in the United Kingdom, where all wild rabbit populations that were sampled had seropositive individuals, in the absence of disease-associated mortality [14, 15]. This simple model can explain the very high mean and large range of seroprevalence in the absence of mortality that has been found in the United Kingdom, and the limited impact of acute infection, following its introduction from mainland Europe [33]. There may be numerous factors underlying the high mean seroprevalence (64%) and large range (10–100%) in the United Kingdom and the failure of RHDV to cause mortality on the scale seen elsewhere, except in a few cases [33]. Current data allow only the examination of two possibilities: differences in contact rates within populations, or differences in host demography. Both of these could explain the differences in seroprevalence due to chronic infection, but differences in contact rates could not explain why some populations are at risk of acute phase invasion and others are not. Changing the contact rate does not affect the ratio of the R_0s of the two phases of infection: with reference to Figure 2, changes in contact rate move the system along a straight line passing through the origin. Since this line does not cross the boundary where $R_{0,A} = R_{0,C}$, changes in contact rate cannot explain how the behaviour of the system would vary amongst populations at $(K, 0, C^*)$, with some potentially suffering mortality due to acute phase invasion, and others not.

Host demography can explain both the differences in seroprevalence amongst populations in the United Kingdom and why some populations are protected from acute-phase-induced mortality, whilst others are not. The differences in the infectious periods of the acute and chronic phases mean that their basic reproductive rates differ markedly in sensitivity to host population dynamics, with $R_{0,A}$ being almost invariant and $R_{0,C}$ being highly variable. The UK range of natural death rates, 0·00289–0·0171, corresponds to $R_{0,A}$ range 1·80–1·86, and $R_{0,C}$ range 1·40–8·26. Where a population is at the chronic-phase-only $(K, 0, C^*)$ equilibrium, its seroprevalence indicates whether it is at risk from acute-phase invasion, and whether such an invasion would lead to depression of the host population, or its extinction. This is because seroprevalence in the chronic-phase-only $(K, 0, C^*)$ equilibrium indicates host natural death rate, b, which determines which phase of infection has the higher R_0 and whether acute infection causes host extinction in the coexistence equilibrium (Fig. 3). The north of the United Kingdom has higher mean seroprevalence and has had fewer lethal outbreaks of RHDV than the south [33]. In the context of the model, this can be explained by lower productivity, therefore lower natural death rates, in the north. In those populations, $R_{0,A} < R_{0,C}$, so acute infection cannot invade the chronic-phase-only equilibrium $(K, 0, C^*)$. In some UK populations, predominantly in the south, $R_{0,A} > R_{0,C}$, making the chronic-phase-only equilibrium $(K, 0, C^*)$ prone to invasion.

Host demography may explain not only differences *within* the United Kingdom but also the difference *between* (most of) the United Kingdom and the regions of mainland Europe where RHDV has had a major impact. In most of the United Kingdom, relatively low natural death rates favour the chronic phase, resulting in widespread, usually high, seroprevalence without mortality. In mainland Europe, where RHDV has had a much more significant impact, recorded seroprevalences were much lower than the United Kingdom, which, we suggest, reflects higher productivity rates, due to a longer breeding season [38] and lower nestling mortality. Thus $R_{0,A} > R_{0,C}$, making the chronic-phase-only equilibrium $(K, 0, C^*)$ prone to invasion. In fact, in parts of mainland Europe, the chronic-phase-only equilibrium may not be relevant due to high natural death rates resulting in $R_{0,C} < 1$.

This analysis is based on equilibrium behaviour of the model, but short-term dynamics also may be important. Firstly, naive populations may suffer significant transient mortality due to the rapid spread of acute infection, before recovering, partially or completely, as illustrated in Figure 4(*a*) and (*b*). Secondly, seasonal population dynamics may be important

in prompting disease re-emergence. Rabbit populations in most of the world are highly seasonal, due to seasonal breeding with high fecundity and high juvenile mortality. In the United Kingdom typically there is a 3- to 4-fold increase in numbers from the winter minimum to the mid-summer peak [24], and 'peaks' in Australia can be even higher.

The rapid appearance of susceptibles during the breeding season would stimulate an increase in the prevalence of acute infection (if it were present in the population), with a consequent increase in mortality, due to its transmission parameter being much greater than that of the chronic phase. With reference to Model Analysis section on page 670, above, an increase in the susceptible proportion, S/H, causes $\mathrm{d}\rho/\mathrm{d}t$ to become positive. Thus in populations where there is coexistence between acute and chronic infection, acute infection would be present throughout the year, at low prevalence and so causing relatively little mortality, except during (and just after) the breeding season when its rapid spread causes large-scale, noticeable mortality. Additionally, in some populations where the chronic phase ultimately excludes the acute, if acute infection were introduced from elsewhere at this 'vulnerable' time then there could be significant transient mortality before the acute phase is excluded. This latter scenario was a finding of our previous modelling work (which assumed that there were distinct pathogenic and non-pathogenic strains of RHDV, rather than acute and chronic phases of infection caused by a single strain) [33]. In practice, these two different circumstances would be difficult to distinguish in the field. Note that this 'seasonal re-emergence' can occur simply a consequence of the transmission dynamics of the two phases, without requiring any physiological change in the host or virus, such as the virus in a chronically-infected host 'switching' to acute infection. More detailed analysis of seasonality is beyond the scope of this paper.

RHDV is a good model system for obtaining new insights into the physiological mechanisms and evolutionary aspects of virulence, since the virus apparently possesses both virulent and avirulent modes of behaviour. Several further studies are needed, including extension of the work of Moss et al. [17] to use whole genome sequences, but with a 7·5 kb genome [16, 18] this is a substantial undertaking. Additionally there needs to be more investigation of the physiological interaction between RHDV and its host. Key assumptions of the model presented here are that chronic infection is (i) infectious, (ii) causes chronic infection upon transmission (due to low-level viral shedding) and (iii) lasts for life (or at least a long period). Although the observational evidence is strong, these assumptions need to be confirmed by experimental studies. To date, the effect of low-dose inoculation with RHDV has not been studied, with laboratory studies using high doses of RHDV (typically 10^3–10^6 LD_{50} [3, 18, 37]), to ensure that (acute) infection occurs.

More generally, the mechanism of infectious chronic infection may be employed by other pathogens that persist at relatively high prevalence in their host species despite their potential virulence, such as *M. bovis* in badgers. A number of pathogens that cause chronic infection are able to 'switch' between a non-infectious latent state and an 'active' infectious state, when conditions may be more favourable to transmission. Latency is a mechanism by which pathogens causing (re-)emerging diseases are able to persist, unnoticed, in between epidemic outbreaks, sometimes for long periods. In the case of RHDV it is not known if an analogous 'activation' of chronic infection may occur, with bouts of increased viral shedding causing acute (rather than chronic) infections upon transmission. (Indeed we have discussed how disease re-emergence could occur simply as a result of seasonal host population dynamics coupled with coexistence between acute and chronic infection, without requiring such a 'switch' in the behaviour of the virus within the chronically infected host.) However, such a mechanism would allow RHDV – and other virulent pathogens whose 'latent' state may actually be infectious and cause chronic infection upon transmission – to spread 'silently', without causing apparent disease. This would increase the likelihood of disease (re-)emergence because there would be more individuals with chronic infection, which may 'activate' in a stochastic event (perhaps due to immunosuppression caused by the stress of crowding or reproduction or another disease), and begin shedding virus at higher levels, causing acute infections upon transmission and initiating disease emergence.

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APPENDICES

(*a*) Estimation of natural death rate, *b*, from population dynamics data

Published demographic studies commonly quote productivity and adult mortality data. In order to estimate the mean natural death rate for UK rabbit populations, for use in a model without age structure, the following approach is used. We consider a disease-free, non-seasonal equilibrium population, at carrying capacity, which therefore has a stable size and age-structure. This population can be represented by an age-structured model and a non-age-structured model. In the age-structured model the population is divided into juveniles, J, and adults, Ad, with the former maturing at rate m. The rate at which new juveniles enter the population is a function of the adult population size, Ad, and productivity rate per adult, p, which is the product of the birth rate per adult and nestling survival. The daily death rate for juveniles is b_J and for adults, b_{Ad}. In the non-age-structured model, the population is of size R and the mean death rate, b, is equal to the mean productivity rate at carrying capacity, because in a stable population each birth has a corresponding death.

The equations of the age-structured model are:

$$\frac{\mathrm{d}J}{\mathrm{d}t} = pAd - (b_J + m)J,$$

$$\frac{\mathrm{d}Ad}{\mathrm{d}t} = mJ - b_{Ad}Ad,$$

$$\frac{\mathrm{d}(J+Ad)}{\mathrm{d}t} = pAd - b_J J - b_{Ad}Ad.$$

For the age-structured and non-age-structured population models to be equivalent, total population sizes must be equal (i.e. $J+Ad=R$) and the 'total population' death rates must be equal (i.e. $b_J J + b_{Ad}Ad = bR$). Solving the models at equilibrium gives $b = pm/(b_{Ad}+m)$.

UK birth rates of 14–22 young per adult female per annum [27], combined with nestling survival in the range 25–75% [24] gives a productivity range of 1·75–8·25 juveniles weaned per adult p.a., consistent with Bell and Webb [23]. Adult annual mortality ranges from 40–80%, giving b_{Ad} values in the range 0·00142–0·00447. The typical age of first successful breeding in the United Kingdom is 9 months (i.e. in the following breeding season), which is 8 months post-weaning, giving a daily maturation rate, m, of 0·00417. With the constraint that $b_J > b_{Ad}$, estimated values of b are in the range 0·00289–0·0171. The typical UK rabbit population has adult mortality of 60% per annum, with 5 juveniles weaned per adult p.a., resulting in $b = 0{\cdot}00862$.

(*b*) Invasion of the uninfected equilibrium (K, 0, 0): calculation of R_0 for each phase

The relevant Jacobian is,

$$J_{(K,0,0)} = \begin{bmatrix} -r & -\alpha & 0 \\ 0 & \beta_1 - d & 0 \\ 0 & \sigma & \beta_2 - b \end{bmatrix}.$$

The eigenvalues satisfy,

$$\lambda_1 = -r, \quad \lambda_2 = (\beta_1 - d), \quad \lambda_3 = (\beta_2 - b).$$

The uninfected equilibrium is unstable (i.e. infection can spread successfully in the population) if the dominant eigenvalue does not have negative real parts. For the uninfected equilibrium to be relevant, λ_1 must be negative, so infection can invade if $\lambda_2 > 1$ (i.e. $\beta_1/d > 1$) and/or $\lambda_3 > 1$ (i.e. $\beta_2/b > 1$). Thus each phase of infection can be considered to have its own R_0, where $R_{0,A} = \beta_1/d$ and $R_{0,C} = \beta_2/b$.

(*c*) Invasion of the chronic-phase-only equilibrium (K, 0, C^*)

The relevant Jacobian is:

$$J_{(K,0,C*)} = \begin{bmatrix} -r & -\alpha & 0 \\ 0 & \dfrac{b\beta_1}{\beta_2} - d & 0 \\ \dfrac{(\beta_2 - b)^2}{\beta_2} & b - \beta_2 + \sigma & b - \beta_2 \end{bmatrix}.$$

The eigenvalues satisfy,

$$\lambda_1 = -r, \quad \lambda_2 = \left(\frac{b\beta_1}{\beta_2} - d\right), \quad \lambda_3 = (b - \beta_2),$$

λ_1 must be negative and a condition for the relevance of (K, 0, C^*) is that λ_3 is negative, so the only eigenvalue that may not have negative real parts is λ_2. Thus acute infection can invade (K, 0, C^*) where $\beta_1/d > \beta_2/b$, i.e. where $R_{0,A} > R_{0,C}$. The system then moves to (H^*, A^*, C^*) or host extinction.

(*d*) Relationship between β_2 and seroprevalence in the chronic-phase-only equilibrium (K, 0, C^*)

At (K, 0, C^*), $H^* = S^* + 0 + C^*$. Seroprevalence $= C^*/H^* = 1 - (S^*/H^*) = 1 - (b/\beta_2)$. Rearranging, $\beta_2 = b/(1 - \text{seroprevalence})$.

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